Claims

What is claimed is:

1. A multifunctional fusion protein comprising a polypeptide chain defining an immunoglobulin region (Ig), a first cytokine (C1) and a second, different cytokine (C2).

2. The fusion protein of claim 1, wherein said Ig, said C1 and said C2 are arranged in an N- to C- terminal direction to produce a fusion protein defined by a formula selected from the group consisting of:

   (i) Ig – C1 – C2;
   (ii) C1 – Ig – C2; and
   (iii) C1 – C2 – Ig,

   wherein a dash represents a polypeptide bond or a polypeptide linker.

3 The fusion protein of claim 1, wherein said C1 or said C2 comprise IL-2, IL-4 or GM-CSF.

4. The fusion protein of claim 1, wherein said C1 or said C2 comprise a subunit of a heterodimeric cytokine.

5. The fusion protein of claim 1, wherein the C1 is a chemokine.

6. The fusion protein of claim 4, wherein said subunit comprises a p35 subunit of IL-12 or a p40 subunit of IL-12.

7. The fusion protein of claim 1, wherein said C1 or said C2 is a human cytokine.

8. The fusion protein of claim 1, wherein said Ig comprises an immunoglobulin heavy chain variable region domain (V_H).

9. The fusion protein of claim 1 or 8, wherein said Ig comprises an immunoglobulin heavy chain constant region.

10. The fusion protein of claim 9, wherein said constant region comprises a hinge region domain, a CH2 domain and a CH3 domain.

11. The fusion protein of claim 10, wherein said constant region further comprises a CH1 domain.

12. The fusion protein of claim 9, wherein said V_H is immunologically reactive with a cancer-specific antigen or a viral antigen.

13. The fusion protein of claim 1, wherein said C1 and said C2, when linked in said fusion protein, have similar circulating half lives *in vivo*.

14. The fusion protein of claim 1, wherein said Ig, said C1, and said C2 are all active under the same conditions.

15. A multifunctional protein complex comprising

a first polypeptide chain defining an immunoglobulin region and a first portion of a first cytokine, and

a second polypeptide chain defining a second cytokine and a second portion of the first cytokine.

16. The multifunctional protein complex of claim 15, wherein the first polypeptide chain is covalently bonded to the second polypeptide chain.

17. The multifunctional protein complex of claim 15, wherein the first cytokine is a dimeric cytokine.

18. The multifunctional protein complex of claim 15, wherein the first cytokine is IL-12.

19. The multifunctional protein complex of claim 15, wherein the second cytokine is IL-2.

20. A multifunctional protein complex comprising at least:

a first fusion protein comprising a first cytokine fused to at least a portion of an immunoglobulin light chain, and

a second fusion protein comprising a second, different cytokine fused to at least a portion of an immunoglobulin heavy chain.

21. The protein complex of claim 20, wherein the first fusion protein is covalently bonded to the second fusion protein.

22. The protein complex of claim 21, wherein the portion of the immunoglobulin light chain is disulfide-bonded to the portion of the immunoglobulin heavy chain.

23. The protein complex of claim 20, wherein the amino-terminus of the first cytokine is fused to the carboxy-terminus of the immunoglobulin light chain.

24. The protein complex of claim 20, wherein the amino-terminus of the second cytokine is fused to the carboxy-terminus of the immunoglobulin heavy chain.

25. The protein complex of claim 20, wherein the first cytokine is IL-12.

26. The protein complex of claim 20, wherein the second cytokine is IL-12.

27. A multifunctional fusion protein comprising a polypeptide chain defining a first cytokine (C1) and a second, different cytokine (C2), wherein said C2 when free has an circulating half life *in vivo* greater than about twice that of free C1, but when said C1 is linked to said C2 in said fusion protein said C1 has a circulating half life *in vivo* about the same as said C2 in said fusion protein.

28. The fusion protein of claim 27, wherein said C1 is IL-2 or GM-CSF.

29. The fusion protein of claim 27, wherein said C2 is IL-4, IL-12, or a subunit thereof.

30. The fusion protein of claim 27, wherein a C-terminal end of said C1 is linked to an N-terminal end of said C2.

31. The fusion protein of claim 27, wherein a C-terminal end of said C2 is linked to an N-terminal end of said C1.

32. The fusion protein of claim 30 or 31, wherein said C-terminal end is linked via a polypeptide linker to said N-terminal end.

33. The fusion protein of claim 32, further comprising an immunoglobulin region (Ig).

34. The fusion protein of claim 33, wherein said Ig comprises an immunoglobulin heavy chain variable region domain (V_H).

35. The fusion protein of claim 33, wherein said Ig comprises an immunoglobulin heavy chain constant region.

36. The fusion protein of claim 35, wherein said constant region comprises a hinge region domain, a CH2 domain and a CH3 domain.

37. The fusion protein of claim 36, wherein said heavy chain constant region further comprises a CH1 domain.

38. The fusion protein of claim 27, wherein said free C2 has a circulating half life *in vivo* at least about 4-fold greater than that of C1.

39. The fusion protein of claim 38, wherein said C2 has a circulating half life about 8 times greater than that of C1.

40. A nucleic acid encoding the fusion protein of claim 1, 15, 20, or 27.

41. A cell comprising the nucleic acid of claim 40.

42. A method of preparing a fusion protein comprising a first cytokine (C1), a second, different cytokine (C2), and a targeting moiety capable of targeting a preselected locus in a mammal, the method comprising the steps of:

(a) expressing in a host cell a nucleic acid encoding a fusion protein comprising the C1, the C2 and the targeting moiety capable of targeting said fusion protein to the preselected locus when said fusion protein is administered to the mammal; and

(b) harvesting said fusion protein.

43. A method of targeting a first cytokine (C1) and a second, different cytokine (C2) to a preselected locus in a mammal, the method comprising:

administering to the mammal a multifunctional fusion protein comprising a C1, a C2, and an immunoglobulin region (Ig) capable of targeting said fusion protein to the preselected locus in the mammal.

44. The method of claim 42 or 43, wherein said Ig comprises an immunoglobulin heavy chain variable region domain immunologically reactive with an antigen disposed at the preselected locus in the mammal.

45. The method of claim 44, wherein said antigen is a cancer-specific antigen or a viral antigen.

46. The method of claim 42 or 43, wherein said Ig comprises an immunoglobulin heavy chain constant region capable of binding an immunoglobulin Fc receptor disposed at the preselected locus in the mammal.

47. The method of claim 42 or 43, wherein said Ig, said C1 and said C2 are all active when said fusion protein is administered to the mammal.

48. The method of claim 42 or 43, wherein said mammal is a human.

49. The method of claim 42 or 43, wherein said C1 is IL-12 or a subunit thereof.

56. The method of claim 49, wherein said C2 is selected from the group consisting of IL-2 and GM-CSF.

57. A method of treating disease in a mammal, the method comprising administering to the mammal the protein of claim 1, 15, 20, or 27.

58. A method of treating disease in a mammal, the method comprising administering to the mammal the nucleic acid of claim 40.

59. A method of treating disease in a mammal, the method comprising administering to the mammal the cell of claim 41.